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Variability in larval settlement of abalone on artificial collectors

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Abstract

The spatial and temporal variability in settlement of greenlip abalone, *H. laevigata* on artificial settlement collectors was examined over three years. Pairs of the collectors of an improved design were deployed on the sea bed at six sites over a number of different spatial scales. A period of peak settlement occurred between October and December in each of the three years at all sites. The similarity in size frequencies between sites at the settlement peaks demonstrated that spawning was epidemic, occurring over a period of 2–3 days. The synchronous timing and intensity of peaks at four sites within 2 km of each other contrasted with that of a site 8 km distant. Adult densities between sites during the spawning season ranged from 0.0 m^{-2} to 1.2 m^{-2}. Over this range of densities, numbers of larval settlers were independent of local adult abundance.

Collectors are a useful tool in examining inter-annual variability in the timing and synchrony of spawning and in measuring the relative abundance of abalone larvae. Growth rates inferred from "cohorts" followed at two week intervals allowed settlement/spawning times to be more accurately estimated than with previous methods such as gonad indices.

Keywords: *H. laevigata*, larval settlement, larval collectors, spawning, epidemic spawning.

Introduction

Artificial collectors have been successful in measuring the intensity of larval settlement in many marine groups, including lobster (Booth and Tarring 1986, Phillips 1986), echinoderms (Harrold *et al.* 1991, Keesing *et al.* 1993), scallops (Sause *et al.* 1987) and abalone (Keesing *et al.* 1995, Nash *et al.* 1995). Estimation of the relative abundance of larvae on both a temporal and spatial scale is important for determining inter-annual variation of settlement and to establish the scale of larval dispersal in order to begin to understand metapopulation structures in the area.

Keesing *et al.* (1995) used a modified abalone larval collector based on that used by Nash *et al.* (1995) and highlighted several problems associated with the laserlight plate collector design. The high sediment and degree of algal fouling after four weeks made sorting the samples time consuming, the surface area of the collector was simple and limited and the collector had minimal trapping capabilities. The development and field use of a novel collector design, incorporating a variety of surface orientations and light regimes that may be important to settling larvae, is described in this paper. The collector must be able to capture enough abalone larvae to give enough power to detect any patterns in settlement. Using the new design, questions relating to temporal and spatial variability in larval settlement can be addressed.

South Australia has five species of abalone, *Haliotis laevigata* Donovan, *H. rubra* Leach, *H. cyclobates* Peron and Lesueur, *H. roei* Gray and *H. scalaris* Leach. Only the first two are commercially fished. This study set out to determine the temporal and spatial variability in larval

greenlip abalone (*H. laevigata)* settlement at six sites. We compared the larval abundance at sites of high and low adult abundance on Taylor Island and McLaren Point. Temporal variation was measured by comparing larval settlement at several sites over a three year period. By deploying collectors in the field from August to February some years, we were also able to determine blacklip (*H. rubra*) settlement.

Materials and Methods

Collectors

At the original site used by Keesing *et al.* 1995 (McLaren Point, South Australia), we compared the larval collecting ability of three artificial collector designs, including the original laserlight plate design (type 1– see Keesing *et al.* 1995). Collector type 2 was constructed of 13 layers of a black polycarbonate material which had a complex surface area (3 m^2) and a volume of 0.3m^3 (Fig.1). The surface of neighbouring sheets formed channels which provided a variety of surface orientations and an enhanced trapping capability. It also provided a variety of light regimes to incorporate any larval preference. Type 3 was constructed of 10 layers of fluorescent diffuser material held separate with a central rod which directed the collector into the current.

Study sites

Once comparisons were made between collectors, the most efficient collector design was chosen and used to establish spatial variation in larval settlement at five sites in Thorny Passage, near Port Lincoln. In addition, the relationship between adult abundance and the number of larval abalone

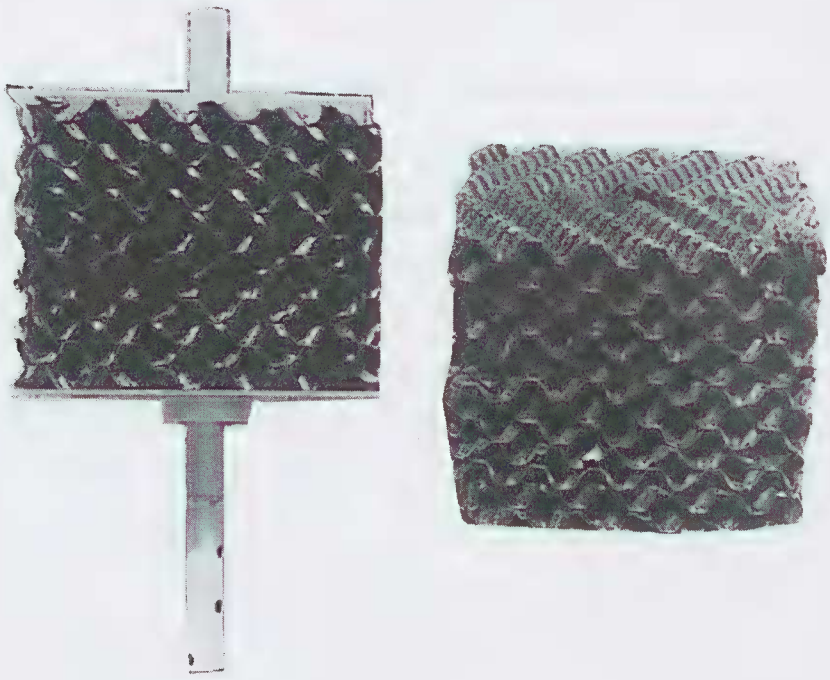

Figure 1. Collector design 2 used in this study.

settling was compared by choosing sites of varying adult abundance. Four of the sites (two high and two low adult abalone abundance) were located 2 km apart on Taylor Island (136°1'00", 34°54'00"). An additional site (Passage site) with no known adult population of *H. laevigata* within 500m (P. Clarkson pers. comm.) was set 2 km west of Taylor Island. The remaining site (McLaren Point – 136°1'00", 34°47'90") with a high adult abundance, was 8 km north of Taylor Island (Fig.2).

At each site, six collectors were placed one metre above the seabed. The collectors were fixed in pairs to a solid yoke attached to a cement-filled tyre. During the experimental period we changed the design of the yoke from PVC piping to a flat mild steel. This was especially useful in areas of high water movement where the PVC piping was prone to break.

The distance between pairs of collectors varied from 2–6 m. The collectors were placed at a water depth of 5–6 m at four sites and at 11–12 m at the remaining two sites. Five of the sites were on the sand-reef interface near an adult greenlip abalone population and the remaining site (Passage site) was on a sandy bottom.

The collectors were conditioned naturally in the sea with diatoms and bacteria which created a

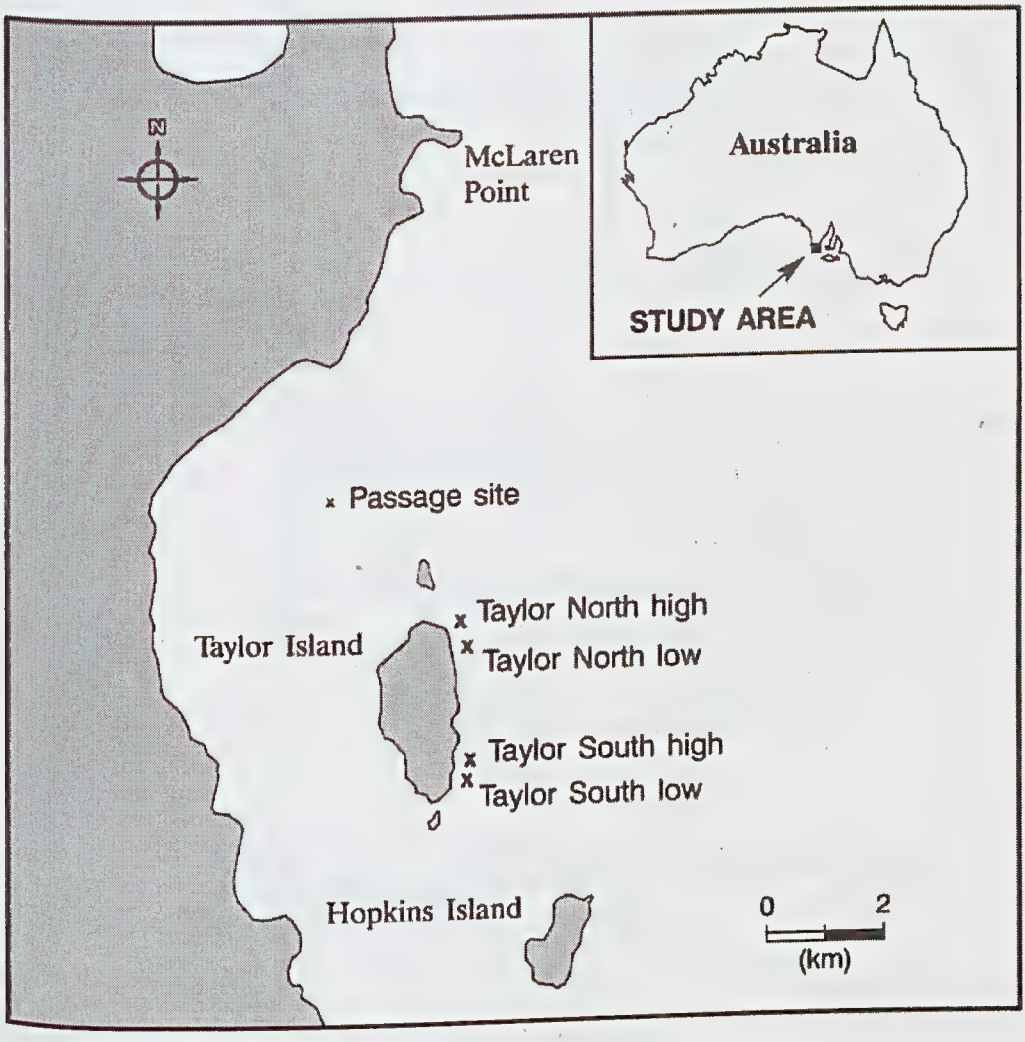

Figure 2. Study site locations in Thorny Passage, near Port Lincoln, S.A.

Figure 3. Total number of post-larvae in the collectors at the Taylor South high site from 1993–96 and the mean gonad indices for greenlip (*H. laevigata*) and blacklip (*H.rubra*) at the same site over the same period. Vertical bars are the standard errors.

suitable substratum for settlement and growth of juvenile abalone (Nash *et al.* 1995, R.Grove- Jones pers. comm.).

Collectors were monitored for five months over the summer spawning season at five sites and all year round at one site (Taylor South high). Deployment times of between two and four weeks were used according to Keesing *et al.* (1995). Two replicate pairs of collectors were picked up every fortnight, each pair representing two and four week collections. The collectors were brought to the surface and placed into tagged bags on the boat. All retrieved collectors were replaced with new collectors. Hence at all times there were two conditioned collectors *in situ* to allow larval settlement to occur. The retrieved collectors were returned to the laboratory and frozen until examination. The collectors were then thawed and washed thoroughly with fresh water to remove any settled abalone.

Scrubbing of the collectors to remove juvenile abalone was unnecessary. In a pilot experiment to determine the benefit of scrubbing the collectors we found that we retrieved only 1.4 % (SE = 0.74, n=6) more abalone. All sediment greater than 125μm was collected and stored in 100% ethanol. Small amounts of the dye Rose Bengal were added to distinguish animal tissue from sand to make sorting easier. Each sample was later examined under a dissecting microscope and the number and size of abalone found were recorded.

Species identification was difficult under a binocular microscope if the post-settlement abalone were less than 500μm shell length (SL). Abalone larger than this (as found on the four week collectors) were able to be identified by comparing shell shape with blacklip and greenlip abalone reared in the laboratory. In order to link the settlement peak with a specific species, we examined all four week collectors around the settlement peak. In addition, gonad indices taken year round gave us some idea of spawning activity among the different species.

Growth rates of juvenile abalone

The growth rate of abalone on the collectors was determined by following the prominent cohorts from two collectors, placed on the seabed on the same date but retrieved two and four weeks later, respectively. A linear growth model was assumed during the first four weeks.

Gonad Indices

Once a month, at the time of retrieval of a collector, 6–10 samples of gonad from some or all of the three most commonly found species at the Taylor South high site (*H. laevigata*, *H. rubra* and *H. cyclobates*) were taken to indicate reproductive condition and hence spawning activity. All individuals were of reproductive age (over 120 mm SL for *H. laevigata* and *H. rubra* and over 40 mm SL for *H. cyclobates*). Samples were stored frozen or fixed in 10% formalin for later examination. Gonad indices were measured by slicing a cross section through the visceral mass at a point midway between the spiral coil and tip. The cross section was then traced onto paper and enlarged 5 times by photocopier and weighed. The gonad portion was weighed relative to the visceral mass (to the nearest 10 mg) and the gonad index determined by the following equation (after Shepherd and Laws 1974):

$$\text{Gonad Index} = \frac{\text{weight of gonad}}{\text{weight of gut + gonad}} \times 100$$

Adult densities

At each of the six sites, two permanent 50 m long transects were marked. Annual summer surveys one metre either side of the transect line recorded the number and size frequency of all greenlip abalone found. From the size frequency data, it was possible to distinguish cohorts relating to distinct age classes to at least four years old (Shepherd *et al.* 1992). The relationship between the density of reproductively active abalone (> 100mm SL) and the density of larvae at the peak settlement on the collectors at each site was tested by ANOVA. The density of adults was assumed to be relatively stable in the area between the time of peak settlement and survey of adults (conducted in November each year).

Taylor South High
shell length (µm)
number
Taylor South Low
shell length (µm)
number
Taylor North High
shell length (µm)
number
Taylor North Low
shell length (µm)
number

Results

Collector type

We examined qualitatively the relative time spent cleaning the collector after retrieval and time spent sorting samples as important factors in choosing a practical design. Unpublished results by the authors show that, comparatively, the black multilayered polycarbonate collector was equally efficient at capturing abalone larvae as the type 1 collector and superior to type 3. The accumulation of algal growth on collector types 1 and 3 over a short exposure time of two weeks increased sorting time. The black polycarbonate sheets (type 2) retained very little algae and therefore reduced sorting time. In addition, the complex surface area of type 2 also provided a variety of light and surface orientations to accommodate any larval preferences. Type 2, the black polycarbonate collector, was therefore chosen as the optimal design for our purposes. At each collector pickup, two replicate samples were taken for each period of deployment (two and four weeks). A linear regression analysis demonstrated no significant difference in the number of abalone found between replicate collectors ($F=251.5$ $r^2=0.79$, $n=70$), thus justifying the utility of the collectors as a quantitative tool.

Larval settlement

The numbers of settlers recovered from the collectors (Fig. 3) and their size composition (Fig.4) indicate that, while there is a low continuous level of settlement throughout the year, short intense periods of settlement occurred at each site. Data are shown for the Taylor South high site where the data set is most complete (Fig. 3). Because the replicates were not significantly different, the data represent the sum of abalone found on the two collectors. At this site collectors were deployed year round in 1994/1995 and over the spring-summer period for the other three years. The peaks are likely to represent a *H. rubra* (August) and a *H. laevigata* (October–December) settlement, as suggested by the decline in the respective gonad indices of the two species (Fig. 3).

The peak *H. laevigata* settlement in 1993 was synchronous at the four Taylor Island sites, and asynchronous with the peak settlement at the site 8 km distant (McLaren Point) (Fig. 5). This spatial asynchrony was repeated in the subsequent years of study (Fig. 6). The first year's data showed peaks of similar intensity at all sites. Although settlement was markedly less at three of the Taylor Island sites in the last two years of study, the peak was still synchronous at all Taylor Island sites, including the Passage site.

The timing of peak settlement of *H. laevigata* at the Taylor Island sites varied over the three year study but always occurred during the late spring or early summer (early December 1993, late October 1994 and mid December 1995).

The modal mean size of post-larvae was 250μm SL on the 2-week collector retrieved on 12 Nov. 1993 and 475μm SL on the 4-week collector retrieved 2 weeks later. This gave a mean growth rate of

Table 1. The relationship between densities (per meter square) of reproductive adult *H. laevigata* and post-larvae on the collectors. Larval density was taken from the peak settlement occurring over summer.

		Taylor South high	Taylor South low	Taylor North low	Taylor North high	Passage	McLaren Point
1993	Adults	1.15	0.37	0.49	0.75	–	0.64
	post-larvae	65.0	43.3	70.0	37.7	–	56.0
1994	Adults	0.51	0.28	0.42	0.66	0.0	0.45
	post-larvae	66.0	–	–	35.0	10.7	34.7
1995	Adults	0.49	–	0.25	0.42	0.0	0.45
	post-larvae	57.0	–	16.7	11.3	35.6	52.7

– indicates that no data was taken

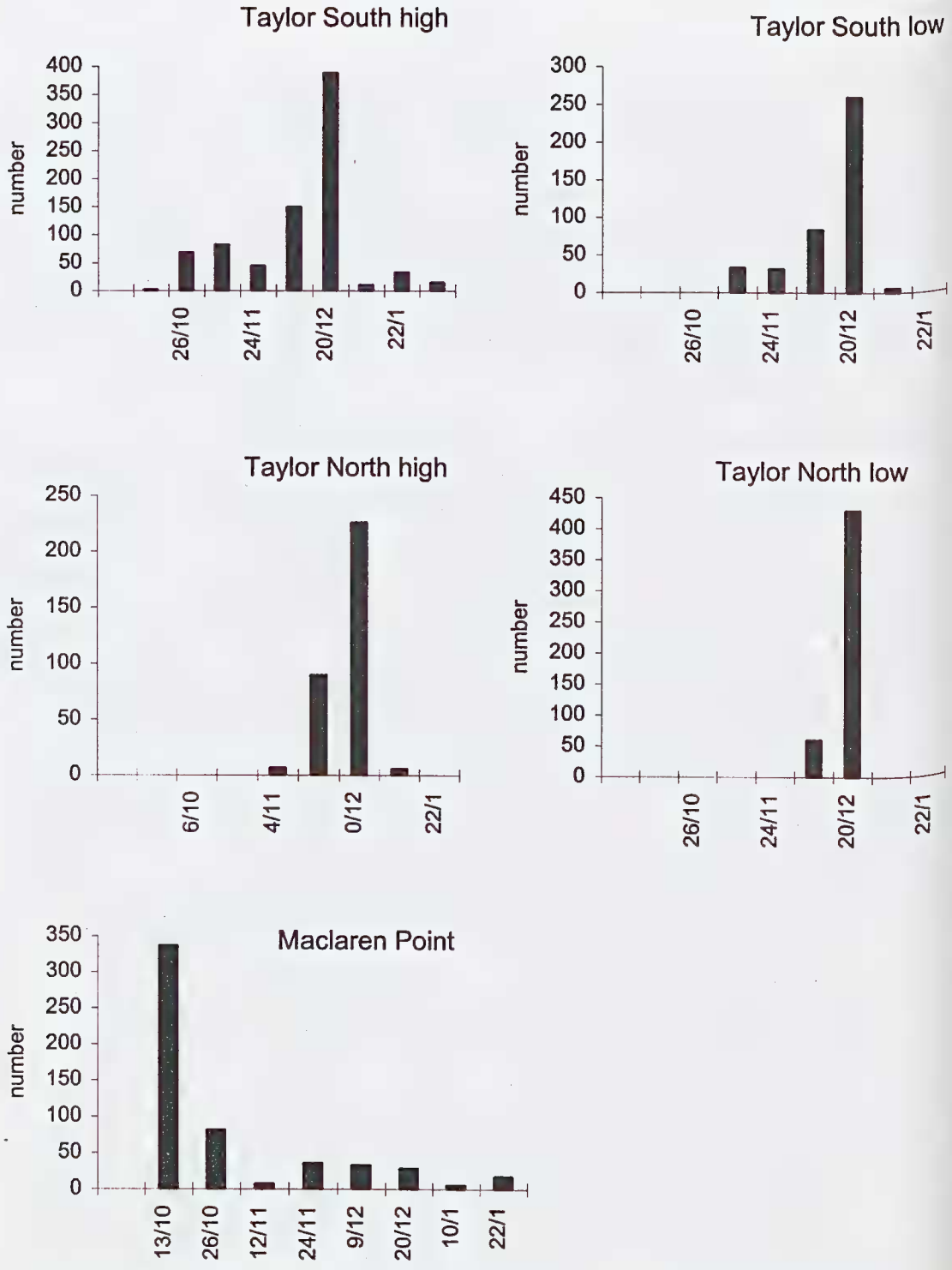

Figure 5. Total number of abalone found on the collectors at all sites in the period October 1993–January 1994.

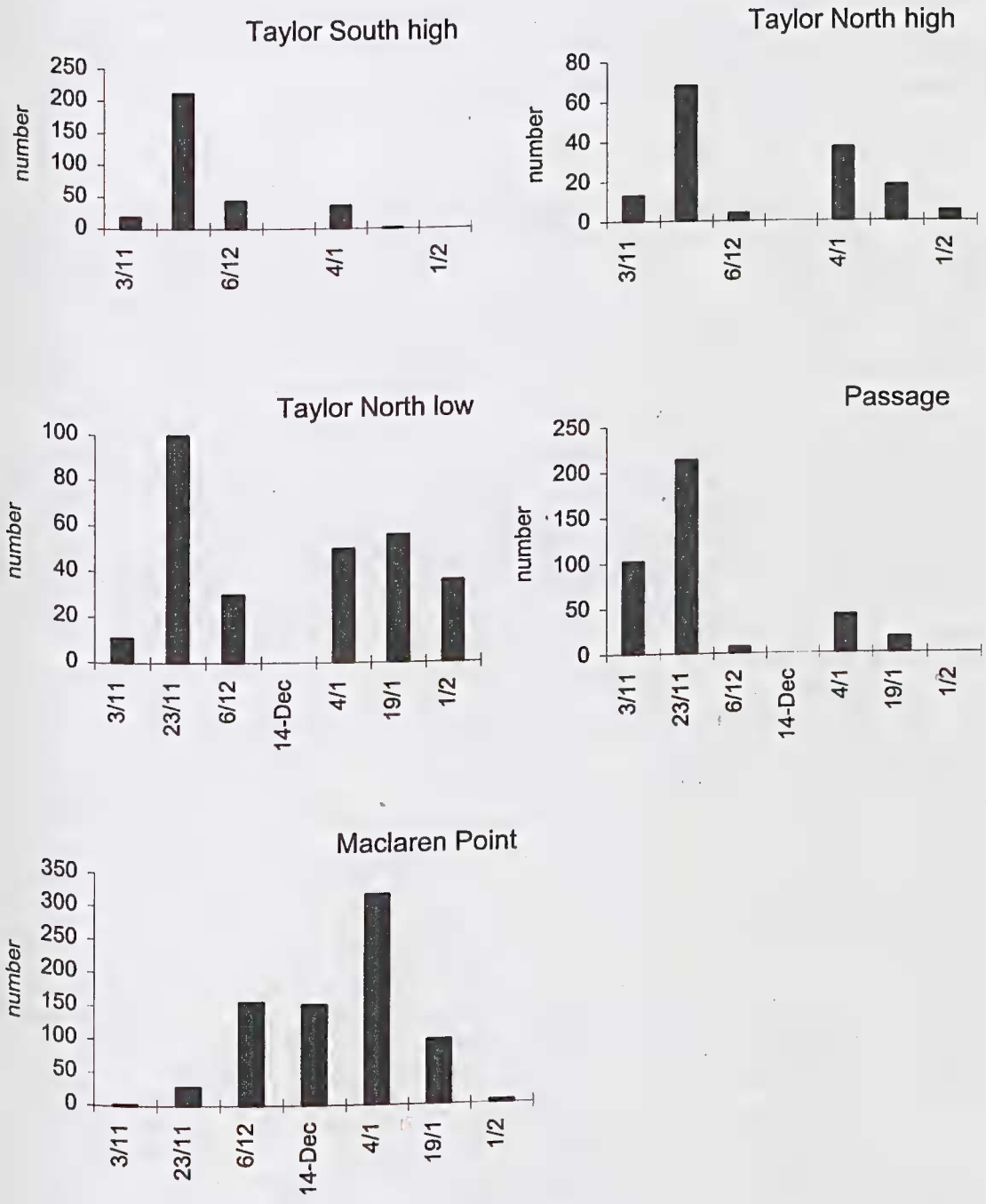

Figure 6. Total number of abalone found on the collectors at all sites in the period November 1995–February 1996.

19μm day^{-1}. Comparable growth rates were found by comparing the size frequency of abalone on several other 2 and 4-week collectors.

Annual surveys over three years produced mean adult densities of 0.66 m^{-2} (SE 0.11, n=6) and 0.36 m^{-2} (SE 0.16, n=5) at the high and low density sites, respectively. At McLaren Point, adult density was 0.51 m^{-2} (SE 0.06, n=3), and at the Passage site, adult density was zero (Table 1). The density of settled larvae at peak settlement ranged between 10.7 and 70.0 m^{-2} and was not related to the density of reproductive greenlip abalone (ANOVA : $F=5.31$, $P<0.001$).

Discussion

The synchronous timing and intensity of the peaks of settlement at the Taylor Island sites within 2 km of each other contrasted with those found at a site 8 km distant. Although our samples represent the cumulative number of surviving settlers over two weeks, the similar size composition of settled abalone collected during the peak of settlement activity (Fig. 4) is indicative of an intense period of settlement perhaps lasting just 2–3 days and suggestive of a epidemic spawning some days before. The low or background density of settlement before and after the peak may represent widely dispersed settlers from a more distant locality or settlers from a local asynchronously spawning parent population.

Were the larvae captured by the collectors conspecific? The reef habitat along Taylor Island supports low to medium density populations of *H. scalaris* and *H. rubra*, and *H. laevigata* can be found at high (Taylor South high, Taylor North high) and low (Taylor South low and Taylor North low) abundances. *H. cyclobates* is common at two sites (Taylor South high, Passage site) while *H. roei* is uncommon at all but one site (McLaren Point). To derive which abalone species were likley to be spawning, we examined gonad indices of the more abundant species present at Taylor South high. With the exception of the 1996/1997 season, the reduction in gonad indices, corresponding with spawning, suggests that spawning episodes of the 4 species occur at different times of the year. *H. scalaris'* peak reproductive activity occurs in March– April, and *H. cyclobates* spawned during winter (1994) and during early to late summer in 1993/1994 and 1995/1996 (unpublished data). *H. roei* is in reproductive condition all year round, but has a major spawning season in April (Keesing *et al.* 1995) and the larvae settle in shallow reef habitat (1–2 m deep) (Shepherd and Laws 1974). It is therefore unlikely that larval settlement of these three species would be recorded on the collectors during the experimental period, or be confused with *H. laevigata* or *H. rubra* settlement peaks.

The differences in settlement recorded between sites 8 km distant may be due to differences in timing of spawning at the two sites. Water temperatures and food availability differ substantially between the two sites and may induce different spawning periodicities. Water temperature and weather variables at the sites are being assessed as environmental cues for spawning in abalone (Rodda and Keesing, in prep.).

The timing of peak settlement in *H. laevigata* at the Taylor Island sites varied slightly over the three year study although it always occurred in late spring or early summer (early December 1993, late October 1994 and mid December 1995). Similarly, at the same site in 1987, Shepherd *et al.* (1992) found that spawning took place in early December. The reproductive condition of the adults is influenced by the available nutrition during the months preceding spawning. The gonad index in *H. laevigata* begins to increase to around 50% by July each year (see Fig. 3), some 4–5 months prior to spawning, peaks at around 75% prior to spawning and tends to fall abruptly after peak settlement.

Some declines in gonad indices were not reflected in settlement peaks. Explanation of such incongruities include: the small gonad sample sizes, which may not accurately reflect the natural range of reproductive activity occurring at the sites, the resorption of gonad material by individuals or a light settlement on the collectors. Changes in periodic gonad indices, as described in this paper, derived from necessarily small sample sizes, are a valuable guide to spawning activity, but are not definitive. At our study sites, for example, they indicated likely differential settlement peaks of *H. rubra* and *H. laevigata* and also excluded the likelihood of larvae of *H. cyclobates* being confounded

with those of *H. laevigata*; *H. cyclobates* spawned 2 months after *H. laevigata* in the 1993/94 season and 2 months before it two years later (Rodda, unpublished data).

The growth rates of about 19µm per day found in this study are similar to that found by Preece *et al.* (1997) for *H. laevigata* in the first month of benthic existence. Because of the short larval period, the growth rates measured on the collectors allow us to backcalculate the date of settlement and spawning more accurately than any other way. Other studies backcalculate settlement from much older juveniles (up to 3 months old) (Sainsbury 1982, Prince *et al.* 1988, Shepherd and Daume 1996), a much cruder method due to the variability in post-larval growth. The ability to backcalculate the probable spawning date is invaluable in understanding the cues that induce spawning in *H. laevigata* (Rodda and Keesing in prep.).

There was no relationship between adult abundance and the number of abalone which settled on the collectors over a 2 km range. The Passage site, located 2 km from Taylor Island, had no adult *H. laevigata* within at least 500m, yet still produced a synchronous settlement peak with the remaining Taylor sites. The population of *H. cyclobates* in the immediate area of the Passage site is an unlikely source of settlers on the collectors, because gonad indices suggest that its spawning activity is in late summer. Shepherd *et al.* (1992) concluded that dispersal along Taylor Island was at least 200m, but our study suggests that the dispersion may be greater. Given tidal currents of 50 cm sec^{-1} or more in Thorny Passage and tidal excursions of several kilometres, larvae could be dispersed on the scale of kilometres. The synchronous peak on the Passage collectors with that of sites on Taylor Island is consistent with this view.

Artificial collectors provide a valuable tool for the study of the early life history of abalone, and associated questions such as spawning cues of adults. Our new design of collector with high capture ability allows comparisons within and between sites over time. Now the way is open to monitor larval availability to abalone habitat, and to answer the more difficult questions posed by Day and Shepherd (1995) namely, the relation between larval availability and recruitment and the factors that influence recruitment success.

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